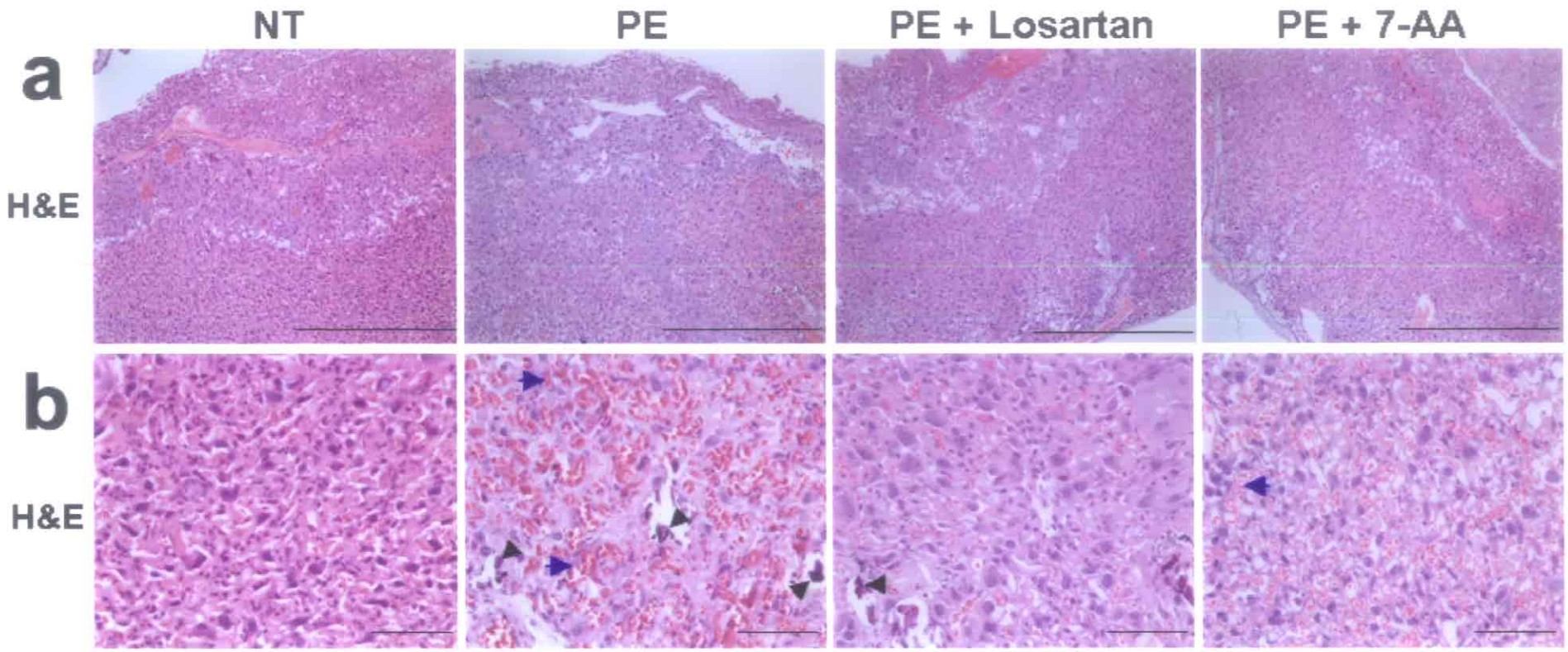
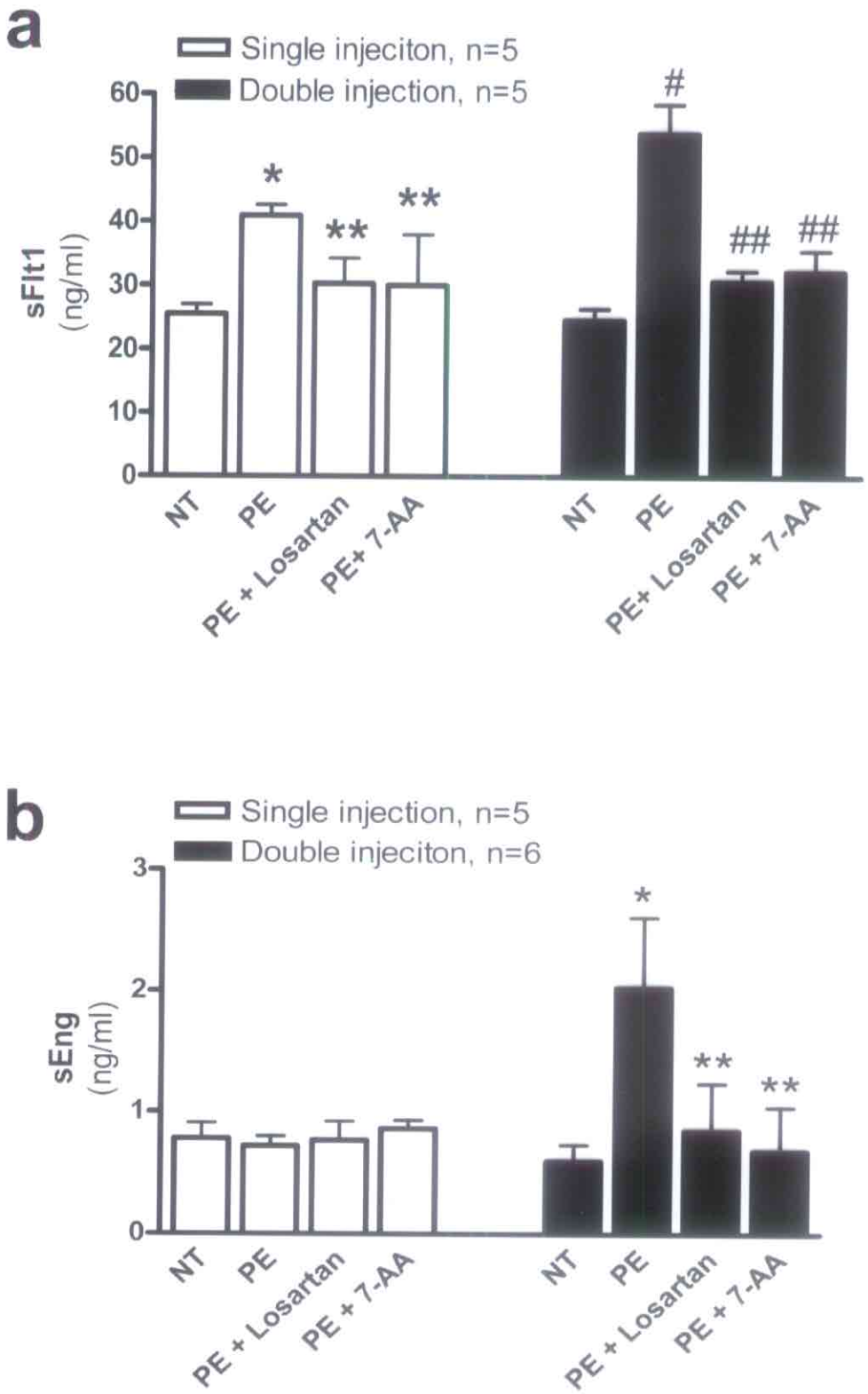
Supplementary Figure S1



Supplemetnary Figure Legend

Supplementary Figure S1. Placental damage induced in mice by injection of IgG from preeclamptic women via AT1 receptor activation. Placentas of pregnant mice after injection of IgG (~ 800 μg) from preeclamptic women showed an increase in placental calcifications (black arrows) and extensive cellular disorganization of the labyrinth zone (blue arrows) as compared to mice injected with IgG from normotensive women. These placental changes were less pronounced when pregnant mice were injected with IgG from preeclamptic women along with Losartan or the 7 AA epitope peptide. Panel a, 4X magnification of all three mouse placental zones: the maternal decidua, junctional zone and the labyrinthine zone (from top to bottom). Panel b, 20X magnification of the labyrinthine zone, where there is maternal-fetal blood exchange. PE, mice injected with IgG from preeclamptic women. NT, mice injected with IgG from normotensive pregnant women.

Supplementary Figure S2



Supplementary Figure S2. Injection of IgG from preeclamptic patients into pregnant mice leads to increased maternal circulating levels of sFlt1 and sEng in a dosage-dependent manner. (a) The level of sFlt1 in blood circulation of pregnant mice was induced by IgG from preeclamptic women in dosage dependent manner. IgG (~ 800 μg per injection) from normotensive or preeclamptic pregnant women was introduced into pregnant mice via retro-orbital injection once at gestation 13 or at gestation days 13 and 14. Plasma was collected at gestation day 18 and the concentration of sFlt1 was determined by ELISA. * $p < 0.05$ versus pregnant mice injected with normotensive IgG. ** $p < 0.01$ versus pregnant mice injected once with preeclamptic IgG. # $p < 0.05$ versus pregnant mice injected twice with normotensive IgG. ## $p < 0.01$ versus pregnant mice injected twice with preeclamptic IgG. (b) An increase in the circulating concentration of soluble endoglin (sEng) required two injections of IgG from preeclamptic women. IgG from normotensive or preeclamptic pregnant women were introduced into pregnant mice via retro-orbital injection once at gestation 13 or at gestation day 13 and 14. Plasma was prepared at gestation day 18 and the concentration of sEng was determined by ELISA. * $p < 0.05$ versus pregnant mice injected twice with normotensive IgG. ** $p < 0.02$ versus pregnant mice injected twice with preeclamptic IgG.

Supplementary Table S1. Injection of IgG from preeclamptic women results in small placentas and fetuses in pregnant mice

	n	NT	PE	PE + 7-AA	PE + Losartan	7-AA	Losartan
Fetal Weight (g)	49	1.28± 0.15	1.01±0.17*	1.21±0.12**	1.10± 0.09**	1.24± 0.18	1.22±0.14
Placental Weight (g)	60	0.125±0.013	0.083±0.015*	0.120±0.012**	0.119±0.020**	0.121±0.012	0.120± 0.014

Pregnant mice were injected with IgG from women with preeclampsia in the presence or absence of losartan or the seven amino acid epitope peptide (7-AA). Pregnant mice injected with IgG from normotensive pregnant women were used as controls. Mouse placental and fetal weights were measured at gestational day 18 when mice were sacrificed. PE, preeclampsia; NT, normotensive; All parameters were measured at gestation day 18. Data are expressed as mean ± SEM. * $P < 0.05$ versus NT IgG treatment; ** $P < 0.05$ versus PE IgG treatment. Fetal weight and placental weight are the average of the litter for each group (7 to 8 pups per litter).

Supplementary Table S2. Clinical Characteristics of the study patients

	Normal (n=25)	Severe Preeclampsia (n=27)
Maternal Age (yrs)	28.4± 6.5	32.3± 6.4
Gestational Age (wks)	38.9± 1.0	32.2± 4.0
Primiparous (%)	33%	81%
Systolic Blood Pressure (mmHg)	< 140	178± 16
Diastolic Blood Pressure (mmHg)	< 90	108± 16
Proteinuria (g protein/g creatinine)	< 0.3	6.9± 1.7
sFlt-1 (ng/ml)	1.7± 0.6	6.0± 1.5
AT1-AA Induced NFAT-luc Activity	1.0± 0.7	7.8± 1.9*

AT_1-AA induced luciferase activity was measured as described under Methods. Data are expressed as mean ± SEM. * $P < 0.05$ versus normotensive IgG.